

Poplar Rust: The *Melampsora larici-populina* Genome Sequencing Project

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The basidiomycete *Melampsora larici-populina* is responsible for the leaf rust disease in *Populus* species. Urediniospore germings of this obligate biotrophic fungus usually penetrate the host plant through stomatal openings, differentiates a series of infection structures in the intercellular space, and exhibits highly localized penetration of the host cell wall to establish a haustorium. Hyphae then proliferate in the leaf parenchyma and produce golden pustules filled with masses of urediniospores on the lower leaf surfaces. *M. larici-populina* causes severe economic losses in European poplar plantations and has recently been detected in Canada. Selection for resistance to this biotrophic pathogen is thus an important challenge for poplar breeders. Severe damages occur through decreased photosynthesis efficiency, early defoliation and increased susceptibility to other pests and diseases.

The PGF is sequencing this organism by the whole genome shotgun (WGS) method. Generally we make genomic libraries containing 3 different insert sizes. For all libraries, the DNA is randomly sheared, fragments end repaired and size selected prior to cloning. After cleanup the 3kb insert is ligated into pUC18 vector and the 8kb and 40kb fragments are ligated into pMCL200 and pcc1Fos vectors. The libraries are sequenced to a depth of approximately 4X, 4X, and 0.5-1X coverage respectively.

The JGI has recently sequenced the genome to approximately 4x coverage of this plant pathogen, and is currently sequencing to the 8-9X depth prior to final assembly. The initial size was estimated to be approximately 61 Million bases (Mb), and analysis at the 4X assembly allowed us to revise this estimate to approximately 95 Mb. Library construction, genome assembly and size analysis will be presented.

This work was performed under the auspices of the US Department of Energy's Office of Science, Biological and Environmental Research Program, and by the University of California, Lawrence Livermore National Laboratory under Contract No. W-7405-Eng-48, Lawrence Berkeley National Laboratory under contract No. DE-AC02-05CH11231 and Los Alamos National Laboratory under contract No. DE-AC52-06NA25396.